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Genetic diversity, phenotypic variation and local adaptation in the alpine landscape: case studies with alpine plant species

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Abstract Plant survival in alpine landscapes is constantly challenged by the harsh and often unpredictable environmental conditions. Steep environmental gradients and patchy distribution of habitats lead to small size and spatial isolation of populations and restrict gene flow. Agricultural land use has further increased the diversity of habitats below and above the treeline. We studied the consequences of the highly structured alpine landscape for evolutionary processes in four study plants: *Epilobium fleischeri*, *Geum reptans*, *Campanula thyrsoides* and *Poa alpina*. The main questions were: (1) How is genetic diversity distributed within and among populations and is it affected by altitude, population size or land use? (2) Do reproductive traits such as allocation to sexual or vegetative reproduction vary with altitude or land use? Furthermore, we studied if seed weight increases with altitude. Within-population genetic diversity of the four species was high and mostly not related to altitude and population size. Nevertheless, genetic differentiation among populations was pronounced and strongly increasing with distance. In *Poa alpina* genetic diversity was affected by land use. Results suggest considerable genetic drift among populations of alpine plants. Reproductive allocation was affected by altitude and land use in *Poa alpina* and by succession in *Geum reptans*. Seed weight was usually higher in alpine species than in related lowland species. We conclude that the evolutionary potential to respond to global change is mostly intact in alpine plants, even at high altitude. Phenotypic variability is shaped by adaptive as well as by random evolutionary processes; moreover plastic responses to growth conditions seem to be crucial for survival of plants in the alpine landscape.

Keywords Altitudinal gradient · Clonal reproduction · Common garden · Gene flow · Genetic drift · Isolation by distance · Microsatellites · RAPD · Seed weight

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Genetische Vielfalt, phänotypische Variabilität und lokale Adaptation in der Alpenlandschaft: Fallstudien mit vier alpinen Pflanzenarten

Zusammenfassung Das Überleben von Pflanzen in der alpinen Landschaft ist durch die harschen Umweltbedingungen eine ständige Herausforderung. Die alpine Landschaft zeichnet sich durch steile Umweltgradienten, fragmentierte Lebensräume und genetisch isolierte, oft kleine Populationen aus. Die landwirtschaftliche Nutzung hat diese kleinräumigen Strukturen noch verstärkt. Wir untersuchten die Auswirkungen der reich strukturierten alpinen Landschaft auf Evolutionsprozesse bei vier Arten: *Epilobium fleischeri*, *Geum reptans*, *Campanula thyrsoides und Poa alpina.* Hauptfragen waren: (1) Ist die genetische Diversität in Populationen abhängig von der Höhe über

Meer, von der Populationsgrösse oder der Landnutzung und wie stark sind Populationen genetisch differenziert? (2) Verändern sich reproduktive Eigenschaften wie die Allokation in die sexuelle bzw. die klonale Reproduktion mit der Höhe und der Landnutzung? Weiter untersuchten wir die Auswirkung der Höhe über Meer auf das Samengewicht. Die genetische Diversität war bei allen vier Arten gross und ändert sich in der Regel nicht mit der Höhe oder der Populationsgrösse. Trotzdem fanden wir eine starke Populationsdifferenzierung, die mit der Distanz in allen Fällen deutlich zunahm. Bei *Poa alpina* wurde die genetische Diversität auch durch die Landnutzung beeinflusst. Genetische Drift war bei allen Arten beträchtlich. Die reproduktive Allokation wurde bei *Geum reptans* durch das Sukzessionsstadium und bei *Poa alpina* durch die Höhe und die Landnutzung beeinflusst. Das Samengewicht nimmt bei vielen alpinen Pflanzenarten im Vergleich mit Arten aus dem Tiefland zu. Wir schlussfolgern, dass die Anpassungsfähigkeit an eine sich ändernde Umwelt bei Alpenpflanzen auch in grosser Höhe intakt ist. Phänotypische Variabilität wird nicht nur durch adaptive, sondern auch durch Zufallsprozesse geformt. Phenotypische Plastizität scheint für das Überleben von Alpenpflanzen eine wichtige Rolle zu spielen.

Introduction

The alpine landscape is characterized by a pronounced spatial heterogeneity of abiotic conditions and high temporal dynamics (Körner 2003). With increasing altitude, plant life is challenged by low temperature, a shorter vegetation period, more snow, and harsher conditions by the rising number of weather-related extreme events. Life cycles of alpine plants are threatened by the high uncertainty whether flowering and fruiting, germination and establishment can be successfully completed (Billings and Mooney 1968; Bliss 1971). Plant populations had to adapt to the spatial isolation of suitable habitats and to frequent disturbances caused by climatic oscillations or soil movements. Due to these particular selective pressures alpine floras are clearly distinct from that of the lowlands (Chapin and Körner 1995). Most noticeable is the decline of annuals with increasing altitude and the high number of long-lived species that rely on clonal reproduction for population dynamics (Hartmann 1957; Stöcklin 1992; Klimeš et al. 1997).

The present flora of the European Alps comprises about 4,000 species (Aeschimann et al. 2004) of which 501 are endemic, i.e. occur only in the Alpine mountain region where they have probably evolved. Glacial history and the pronounced heterogeneity of the alpine landscape are responsible for the frequency of endemic taxa in the Alpine Flora, particularly at highest elevations. Nevertheless, many alpine plant species are geographically widespread and occur over the entire Alpine range (Tribsch and Schönswetter 2003; Aeschimann et al. 2004). Patterns of alpine plant species diversity reflect evolutionary processes from the time when the Alps started to take shape as well as species survival and migration during the glaciations. The origin of the Alpine flora dates back to the Tertiary, 10 to 2 million years before present, when the Alps became colonized by plants from the surroundings (Ozenda 1988). As a consequence of plant immigration during the Tertiary, there are close biogeographical relations between the Alps and that of Central-Asian mountains (Kadereit et al. 2008). The latter were an important source for today's Alpine flora (e.g. *Primula*, *Pedicularis*, *Gentiana*, *Saussurea*). During the climatic fluctuations of the Quaternary, expansions and contractions of the alpine glaciers resulted in repeated and drastic changes of the range of the Alpine flora. Only few plant species could survive on ice-free mountain tops (nunataks) or on steep, ice-free slopes (Stehlik 2000). In the last decade, molecular-genetic studies allowed to better understand this glacial history and to locate areas where these plants had been able to survive the ice ages (Stehlik 2003; Hewitt 2004; Schönswetter et al. 2005). Interestingly, the glacial refuges at the edges of the Alps correspond with long-known floristic diversity hot spots of the Alpine flora. This leads to the interesting question in how far the glacial history of the Alpine flora had also an effect on intra-species genetic diversity and on ecologically relevant differentiations and adaptation processes. Understanding such effects is crucial, because a lower genetic diversity would restrict the adaptability of high-alpine species, e.g. to climate change.

While patterns of species diversity in relation to altitude and biogeographic factors have long been studied and are fairly well understood, this is not the case for patterns of intraspecific genetic and phenotypic variability. From lowland studies, it is known that fragmentation and isolation of habitats affect population size and genetic diversity and are risk factors for population survival (Young et al. 1996; Fischer and Stöcklin 1997; Frankham et al. 2002). Small populations show lower fitness (Leimu et al. 2006) and react sensitively to random fluctuations of their size or to unexpected or rapid directed changes of their environment. Also, small populations fairly often suffer from inbreeding and genetic impoverishment, which further restricts their ability to survive (Willi et al. 2006). Therefore, we might expect that in alpine plants, genetic diversity within populations decreases while differentiation among populations increases with altitude given the increasing degree of spatial isolation and the smaller size of high-altitude populations (Crispo and Hendry 2005, but see Till-Bottraud and Gaudeul 2002 for alternative hypotheses). For species that occur over large parts of the Alps, patterns of

genetic diversity may reflect locations of survival during the glaciations and pathways of re-immigration (Holderegger et al. 2002; Stehlik 2003; Schönswetter et al. 2005).

Neutral molecular markers typically provide no information on phenotypic differences among populations, e.g. on variability in phenotypic traits which may be relevant for survival and may result from selection. To test, if phenotypic differentiation in important life-history traits is present, and to know how much it is based on genetic differences, experiments in the common garden or in the greenhouse under controlled environmental conditions are needed (Turesson 1922; Clausen et al 1940; Galen et al. 1991). Differences in the phenotypes among populations need not necessarily have an adaptive background. They may be caused by drift, or result from historical processes irrelevant for the current survival of a population. Reciprocal transplantations or testing population responses to specific experimental treatments are necessary to test if adaptation occurred (Galloway and Fenster 2000). Phenotypic differences among populations of alpine plants may have developed in response to increasing altitude, to successional changes on glacier forelands or due to land use. As a result of the increasing risk of not completing the life cycle in time, the relative importance of sexual versus clonal reproduction might change. Clonal growth, the ability of vegetative reproduction by rhizomes or aboveground stolons is thought to be one of the most remarkable adaptations to the conditions in cold habitats. Seed weight might be affected by altitude because heavier seeds are more likely to successfully establish in harsh conditions (Westoby et al. 1992).

We studied these issues in four typical alpine plant species: *Campanula thyrsoides*, *Epilobium fleischeri*, *Geum reptans* and *Poa alpina.* Here, we present a summary of these case studies with the intention to understand (1) how genetic diversity is distributed within and among populations and is affected by altitude, population size or land use, (2) to what degree reproductive traits such as allocation to sexual versus clonal reproduction vary with altitude or land use and may be explained by local adaptation, genetic drift or phenotypic plasticity (Bradshaw 1984). Furthermore, we studied if seed weight is increasing with altitude between related species-pairs and among populations of single species.

Materials and methods

Study species

Epilobium fleischeri, *Geum reptans*, and *Campanula thyrsoides* are widespread alpine plant species native to the European Alps (Fig. 1 a–c). *E. fleischeri* and *G. reptans* are characteristic plants of glacier forelands appearing within few years after ice retreat. *C. thyrsoides* is found in mesic subalpine to alpine meadows on calcareous soils. Habitats of the three species are patchily distributed in the Alps with geographic distances among populations of around 5–30 km. The three species differ with respect to several important life-history traits that might influence the partitioning of genetic diversity within and among populations. *E. fleischeri* has a mixed breeding system (Theurillat 1979), while *G. reptans* and *C. thyrsoides* are outcrossing (Rusterholz et al. 1993, Ægisdóttir et al. 2007). *E. fleischeri* and *G. reptans* are long-living, clonal plants with a distinct capacity of horizontal spread. All three species have wind-dispersed seeds and are pollinated by insects. The seeds of *E. fleischeri* are frequently dispersed by wind more than 1 or a few km, while most seeds of *G. reptans* are dispersed within hundreds of meter, the seeds of *C. thyrsoides* are hardly dispersed over more than a few meters (Tackenberg and Stöcklin 2008).

The wind-pollinated *Poa alpina* is common throughout the northern hemisphere in subalpine and alpine pastures and nutrient-rich meadows, but also as a pioneer in scree slopes and in snowbeds. In the European Alps, *P. alpina* is among the most important fodder grasses due to its high content of fats and proteins. Similar to other species in the genus, *P. alpina* constitutes a polyploid complex with frequent aneuploidy and highly variable chromosome numbers. In Switzerland, reported chromosome numbers range from $2n = 22$ to $2n = 46$. Some plants of *P. alpina* produce seeds, while others reproduce vegetatively by forming bulbils instead of seeds. The latter grow into little plantlets on the maternal plants, which are therefore called pseudo-viviparous (Fig. 1d).

Genetic diversity

For the analysis of genetic diversity in *E. fleischeri*, *G. reptans* and *C. thyrsoides*, 20 individuals from each of 20 populations per species were sampled across the central Swiss Alps, with a similar altitudinal range of about 1,500 m. To avoid resampling the same clone in *E. fleischeri* and *G. reptans*, a minimum distance of 4 m between individual samples was respected. Population size was estimated by counting flowering individuals or, when population size exceeded 250, by counting a subset and extrapolating to the occupied area. In the case of *C. thyrsoides*, the sampling was extended to 12 additional populations in order to include populations from the western Alps of Switzerland, presumably belonging to a different area of glacial survival and post-glacial immigration. Information on 47–89 polymorphic loci per species generated with five randomly amplified polymorphic DNA (RAPD) markers in each species were used to assess genetic diversity within populations (Nei's expected heterozygosity H_e, the percentage of polymorphic bands P_p)

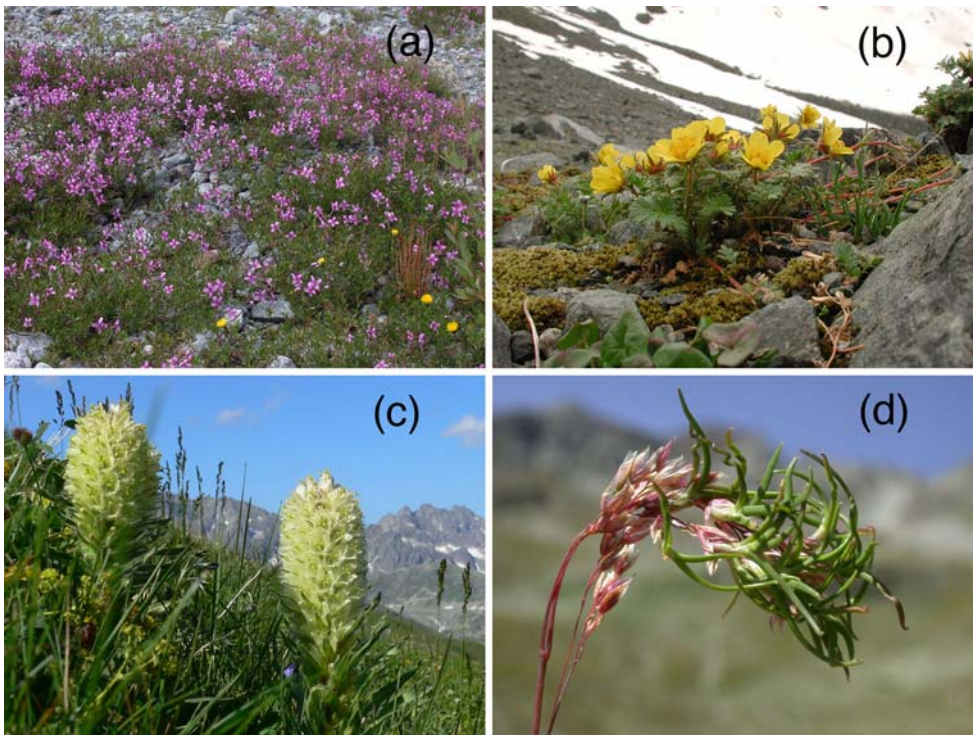

Fig. 1 Study species: **a** Fleischer's willow herb (*Epilobium fleischeri*), **b** Creeping aven (*Geum reptans*), **c** Yellow bell flower (*Campanula thyrsoides*), **d** Alpine meadow grass (*Poa alpina*), vegetatively reproducing by bulbils

as well as genetic differentiation among populations (analysis of molecular variance, Φ_{st}). Within population genetic diversities were correlated to population sizes and altitudes. Spatial patterns of genetic relatedness among populations (isolation by distance) were investigated using Mantels tests correlating the genetic distance matrices (pairwise Φ_{st} values) to geographic distance matrices (Pluess and Stöcklin 2004; Kuss et al. 2008).

Sampling in *P. alpina* for molecular analysis included 569 plants from 54 agriculturally used grassland sites (pastures and meadows) and from 20 natural sites in twelve communities in the Swiss Alps at an altitude from 1,000 to 2,400 m. Using five simple sequence repeat (SSR, microsatellites) markers, we detected 209 bands, between 25 and 61 per locus. Due to polyploidy and frequent aneuploidy in this species, data analysis was based on the presence and absence of SSR bands. We tested whether genetic diversity within populations was related to altitude and land use, and asked whether genetic differentiation among populations was related to geographic distances or to different land use types (Rudmann-Maurer et al. 2007).

Variation in reproductive traits

The relative importance of clonal or sexual reproduction was studied in *Geum reptans* and *Poa alpina*. In *G. reptans*, the same meristem in the axil of leaves can develop either into a flower head or into a stolon. To test the hypothesis that the importance of clonal reproduction, i.e. the number of meristems used for stolon production, increases with altitude and successional age, the relative proportion of sexual versus clonal reproduction was measured in situ in 45 individuals in each of five populations at low and high altitude, and from early and late successional stages on glacier forelands (Weppler and Stöcklin 2005). In a separate glasshouse experiment with this species, we studied the effect of population origin (early vs. late succession) and competition on the relationship between sexual and clonal reproduction using a total of 192 plants from four early and four late successional populations. Here, we were particularly interested to test whether plants had adapted to the different competitive conditions of their respective habitats. In the experiment, this would be shown by an interaction of genetic differentiation (population origin) and the response to competition (Pluess and Stöcklin 2005).

In *P. alpina* two replicates of 690 genotypes from 65 grassland sites, either used as meadows, pastures and from 24 natural habitats were grown in a common garden to test whether plant performance and reproductive traits suggested local adaptation of this species to land use and altitude (Fischer et al. 2008).

Finally, we studied variability in seed weight among 29 related species pairs from low and high altitude and among populations of four species along an altitudinal gradient. We tested the hypothesis that between related species-pairs and among populations of single species a similar trend for increasing seed weight with altitude is present (Pluess et al. 2005).

Results and discussion

Genetic diversity

Within-population genetic diversity in *E. fleischeri*, *G. reptans* and *C. thyrsosides* was similar in all three species (mean H_e between 0.19 and 0.21, Table 1). It was in accordance with other alpine species such as *Eryngium alpinum* (Gadeul et al. 2000), *Trollius europaeus* (Despres et al. 2002) or the average H_e found in the wide range of many other alpine or lowland species (Nybom and Bartish 2004; Till-Bottraud and Gaudeul 2002). Genetic diversity did not depend on population size and was not even reduced in small populations with less than 50 individuals, nor did it decrease with increasing altitude. Other studies also found that genetic diversity does not decrease with altitude (Thiel-Egenter et al. 2009). Generally, changes in genetic diversity were found to be variable along altitudinal gradients as a result of several factors and depending on the elevation at which a species occurs (Ohsawa and Ide 2008).

Genetic differentiation among populations was relatively high (Table 1) and strongly related to geographical distances in all three species, indicating impaired genetic exchanges among populations in the alpine landscape. Within a spatial distance of less than 200 km, pair-wise population genetic differentiation reached maximum values of 29% (Φ_{st}, *C. thyrsoides*) and 44% (Φ_{st}, *E. fleischeri* and *G. reptans*). Even among populations within a distance of only a few kilometres from each other, or among subpopulations on the same glacier foreland, genetic differentiation was present and sometimes high. Obviously, gene flow among populations has a strong random component at all spatial scales. The results suggest that founder effects during the colonization of new sites, and genetic drift play an important role for the genetic structure among populations of alpine species, causing the overall differentiation to be more pronounced than this is usually the case in lowland species.

The amount of genetic differentiation among populations varied in the three insect-pollinated species studied. It was highest in *E. fleischeri*, although the seeds of this plant are spread by the wind up to 100 times farther than those of the other two species (Tackenberg and Stöcklin 2008). Thus, species characteristics other than seed dispersal have to account for this divergence in genetic differentiation. Gene flow due to pollen dispersal as an explanation for the divergence can be excluded, since all three species are pollinated by similar insects: bumblebees, small bees, or flies (personal observation). The measured maximum distance of pollen dispersal, assessed by flower to flower transport of fluorescent powder, amounted to less than 40 m in all three species (Pluess and Stöcklin 2004; Ægisdóttir et al. 2007; J. Stöcklin et al. unpublished). Life form does not provide an explanation either, since all three species are perennial and relatively long-lived, even the non-clonal *C. thyrsoides* (plants usually flower and die at an age of 7–9 years). More likely, the different breeding system may account for the divergence in genetic population differentiation. The flowers of *E. fleischeri*, though being designed for cross-pollination, are self-compatible, while *G. reptans* and *C. thyrsoides* are obligatory outbreeders. Hence self-pollination is possible in *E. fleischeri*, and probably occurs regularly in view of the pronounced clonal growth of this species (Fig. 1a). It is known from numerous studies that self-pollinated species are genetically more differentiated than cross-pollinated ones (Hamrick and Godt 1989; Nybom and Bartish 2004).

In *C. thyrsoides* we found considerable genetic differentiation between the central and the western Alps. Indeed, when populations from the western Alps were added to the above-mentioned analysis, a high proportion of variability (10.3%) was explained by genetic differences between the two groups of populations, and the total genetic variability among populations increased from 16.8 to 27.2% (Kuss et al. 2008). This genetic break line between populations from different bio-geographical regions corresponds well with the presumed post-glacial immigration history from refuge spaces on the edge of the Alps (Stehlik 2000). The same has been observed in other alpine species (Schönswetter et al. 2005; Parisod 2008) and indicates that gene flow among populations since the last ice age has not been strong enough to blur the genetic signature of post-glacial recolonization history in the nuclear genome.

Populations of *P. alpina* also were genetically highly differentiated (>20%), and isolation increased with geographic distance (Rudmann-Maurer et al. 2007). In addition, genetic diversity within populations was positively related to the number of land use types practiced by farmers in a community, indicating the impact of human activity on genetic diversity in alpine plant species. Genetic diversity was higher in pastures than in meadows, which is probably associated with a higher degree of reproduction by seeds in this more heterogeneous land use type. Furthermore, populations from meadows and pastures differed

Table 1 Genetic diversity within populations (H_e Nei's genetic diversity, P_p percentage of polymorphic loci) and differentiation among populations (Φ_{st}, AMOVA-derived fixation index) in three alpine species from the Swiss Alps based on RAPD-molecular markers for 20 individuals in each of 20 populations per species (from Kuss et al. 2008)

	H_e	P_p (%)	Φ_{st} (%)
Epilobium fleischeri	0.19 ± 0.006	74.8 ± 1.4	22.7 ± 0.098
Geum reptans	0.21 ± 0.004	70.1 ± 1.7	14.8 ± 0.028
Campanula thyrsoides	0.20 ± 0.003	61.8 ± 1.3	16.8 ± 0.036

genetically from each other, so that variation in land use also contributed to the genetic differentiation among populations.

Altogether, it can be concluded from these studies and others on genetic diversity (Gugerli et al. 1999; Bingham and Ranker 2000; Thiel-Egenter et al. 2009) that the potential for micro-evolutionary adaptations to environmental change in alpine plants is generally intact. Although the Alpine landscape with its small-scale structures and natural fragmentation restricts gene flow among populations and causes a large spatio-genetic structuring, these effects are not strong enough to severely affect the genetic diversity of alpine plants. This is in contrast to the effects of more recent, man-made habitat fragmentation and population isolation in the lowland. The example of the alpine meadow grass (*P. alpina*) suggests that human land use also influences genetic processes and hence the evolution of alpine plants.

Variation in reproductive traits

Clonal reproduction plays an important role for population growth in *G. reptans*, whose sexual reproduction is not only restricted by a possible lack of pollinators and difficult germination conditions but also by a gall midge that deposits its eggs into the flowers, its larvae impairing the maturation of the ovules (Weppler et al. 2006). Clonal propagation in this plant can easily balance seed losses by predation, at least for local population growth (Weppler and Stöcklin 2006). A study in 20 populations showed that the importance of clonal reproduction, i.e. the proportion of axial meristems developing into stolons, varied greatly among populations in situ (Weppler and Stöcklin 2005). However, the differences could not be explained by the position of a population along an altitudinal gradient, nor by its position in the succession on the glacier foreland (Fig. 2). Rather, populations from marginal locations at highest altitudes and from the lowest sites invested significantly more into clonal reproduction than populations from sites at mean altitudes, where the plant is found most frequently.

A subsequent greenhouse experiment indicated that the reproductive behaviour of populations of *G. reptans* differed in response to competition and according to their origin from different successional stages (Pluess and Stöcklin 2005). Sexual reproduction of plants originating from early successional habitats close to glaciers was more negatively affected by competition than that of plants from older habitats, where competition is naturally more intense, while without competition the behaviour was reversed, plants from early successional habitats performed better than those from late successional habitats (Fig. 3). Altogether, the experimental studies with *G. reptans* suggest

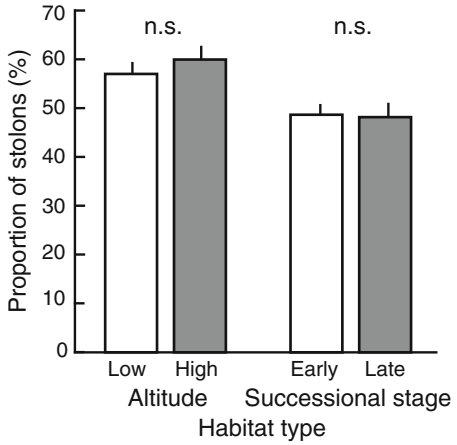

Fig. 2 Frequency (mean ± SE) of clonal reproduction in plants of *Geum reptans* from five populations, each from low and high altitude, and from early and late successional habitats (in total 900 plants). The difference between populations of low + high altitude versus early + late succession is significant (a priori contrast, $P < 0.05$, from Weppler and Stöcklin 2005)

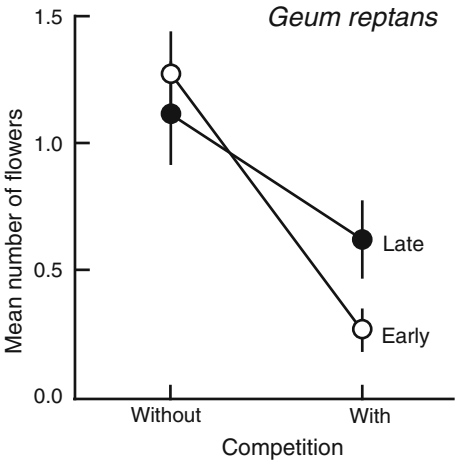

Fig. 3 Number of flowers (mean ± SE) in plants of *Geum reptans* in four populations each from early and late successional habitats (in total 192 plants), grown in a common environment in the greenhouse with and without a competitor ($P = 0.07$ for the interaction competition × habitat origin in ANCOVA, from Pluess and Stöcklin 2005)

some genetic adaptations in the reproductive behaviour of populations from different habitats. However, only part of the observed phenotypic variability could be explained by population origin (successional stage) and the experimental treatment (competition), indicating that drift and/or local environmental conditions play also a role. Moreover, reproductive behaviour in situ as well as in controlled experiments depended strongly on size of the plants: Smaller plants invested more into clonal reproduction, whereas bigger ones more frequently reproduced sexually. This is in line with the hypotheses that seed production is

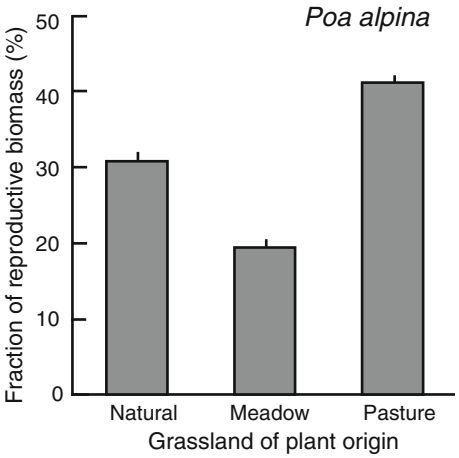

Fig. 4 Fraction (mean ± SE) of reproductive biomass in plants of *Poa alpina* origination from natural, mown, or grazed habitats (in total 1,210 plants from 89 populations), and grown in a common garden in Davos (from Fischer et al. 2008)

more nutrient demanding than the production of clonal offspring, causing smaller plants to invest first in clonal reproduction and larger plants more in seed production (Watson 1984).

In *P. alpina*, a common garden experiment revealed genetic adaptation in the reproductive behaviour to altitude (Fischer et al. 2008). The higher the altitudinal origin of individuals, the more they reproduced vegetatively by bulbils and the less by seeds. This is in line with the hypothesis that, with increasing altitude, clonal reproduction is in advantage compared to reproduction from seeds. The reproductive behaviour of *P. alpina* also depended on the type of land use: plants originating from meadows invested considerably less resources into reproductive structures (seeds and bulbils) than plants from natural habitats, whereas plants from pastures invested much more (Fig. 4). This can be explained by disruptive selection. In meadows, due to heavy competition, increased vegetative growth obviously is of advantage. In pastures, on the other hand, increased reproduction is favoured, since hoof steps of grazing animals create more gaps for offspring establishment from seeds or bulbils. These experimental results match well with the higher genetic diversity of *P. alpina* in pastures as compared with that of plants in meadows, as reported above.

Because of the increasingly difficult conditions for sexual reproduction selection might also affect the size of seeds. Indeed, under harsh environmental conditions in alpine regions, a higher weight of seeds is usually a survival advantage for seedlings, as it means more reserves. We tested this hypothesis in a comparative study with 29 pairs of related plant species, each pair consisting of seed samples from several lowland and alpine populations, respectively. The unit of replication was mean seed size of species pairs (Pluess et al. 2005). Alpine species had on average seeds with a 28% higher weight. This indicates that in contrast to the lowlands, there is a selection pressure at higher altitude favouring species with bigger seeds. If a threshold value of ±25% difference in seed weight was set, 55% of the alpine species had seeds that were heavier, 41% had seeds of equal size and in one case the seed weight of the alpine species was smaller than of the corresponding lowland species. However, when seed weight of numerous (11–34) populations in four species along an altitudinal gradient was studied, no increase in seed weight with increasing altitude was found in any species. Within populations, the variability of seed weight was large, indicating that seed weight has a low heritability (Pluess et al. 2005). Thus, while our results indicate a selection pressure for heavier seeds between related species pairs with altitude, this was not the case at the intraspecific level. The most likely explanation for this finding is that phylogenetic constraints may limit the correlation among altitude and seed weight, operating particularly against selection for larger seeds the closer populations and species are related to each other (Pigliucci 2003).

Conclusions

The above summarized studies suggest that in most alpine plants the potential to adapt to a changing environment is intact. Consequently, alpine plants have the option to react to global change not only by migration, but also by micro-evolutionary adaptation. Such micro-evolutionary processes in situ are facilitated since the topography of the structured alpine landscape rather impedes genetic exchange among populations, so that genetic differences among populations are not levelled out. However, if the environment changes too quickly, local extinction becomes more probable than adaptation. We found plenty of evidence for genetic and phenotypic variation among populations. However, contrary to our expectations, gradual changes of genetic or reproductive traits with altitude were rare, possibly because local differences among habitats of a species are of greater impact than the steady changes along gradients. It is remarkable in this context that the type of human land use, which may change within short distances, can cause genetic differences and cause adaptations in reproductive traits. Plants react to environmental changes also by non-genetic adaptations, and because of their modular construction to a greater extent than this is the case in animals (Bradschaw 1965; Sultan 1987). Our results indicate that such phenotypic plasticity is particularly pronounced in alpine plants. Because of the high heterogeneity of the alpine landscape, the pronounced

capacity of a single genotype to exhibit variable phenotypes is a clear advantage for the persistence and survival of alpine plants.

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